Guidelines for Euthanasia of Rodents Using Carbon Dioxide

Rodents must be euthanized by trained personnel using appropriate technique, equipment and agents. This is necessary to ensure a painless death that satisfies research requirements. Death should be induced as painlessly and quickly as possible. Upon completion of the procedure, death must be confirmed by an appropriate method, such as ascertaining cardiac and respiratory arrest or noting an animal's fixed and dilated pupils (1). Euthanasia should not be performed in the animal room. The euthanasia method must be appropriate to the species, approved in the animal study proposal and conform to the most recent Report of the AVMA Panel on Euthanasia (2).

CO_2 inhalation is the most common method of euthanasia used at NIH for mice, rats, guinea pigs and hamsters. A few important aspects of this procedure are:

1. The euthanasia chamber should allow ready visibility of the animals. Do not overcrowd the chamber: all animals in the chamber must be able to make normal postural adjustments.

2. Compressed CO_2 gas in cylinders is the only recommended source of carbon dioxide as it allows the inflow of gas to the induction chamber to be controlled. Without pre-charging the chamber, place the animal(s) in the chamber and introduce 100% carbon dioxide at the rate of 10-20% of the chamber volume per minute so as to optimize reduction in distress. (For a 10-liter volume chamber, use a flow rate of approximately 1-2 liter(s) per minute.) After the animals become unconscious, the flow rate can be increased to minimize the time to death. Sudden exposure of conscious animals to carbon dioxide concentrations of 70% or greater has been shown to be distressful (3).

3. Animals should be left in the container until clinical death has been ensured. Unintended recovery must be prevented by the use of appropriate CO_2 concentrations and exposure times or by other means.[1]

4. Neonatal animals (up to 10 days of age) are resistant to the effects of CO_2, therefore, alternative methods are recommended (4). Carbon dioxide may be used for narcosis of neonatal animals provided it is followed by another method of euthanasia (e.g. decapitation using sharp blades). Keeping neonates warm during CO_2 exposure may decrease the time to death (5).

5. If an animal is not dead following CO_2 exposure, another approved method of euthanasia (e.g. decapitation) must be added while the animal is under CO_2 narcosis to assure death. Please refer to Appendixes 1 and 2 of the Report of the AVMA Panel on Euthanasia (2) for additional recommended methods.

References

1. NIH Guide for Grants and Contracts. 7/17/2002, notice: OD-02-062. [http://grants.nih.gov/grants/guide/notice-files/NOT-OD-02-062.html]
2. AVMA Panel on Euthanasia. 2000 Report of the AVMA Panel on Euthanasia. J Am Vet Med Assoc 2001, 218:669-696. [http://www.avma.org/resources/euthanasia.pdf]
3. Danneman PJ, Stein S, Walshaw SO. Humane and practical implications of using carbon dioxide mixed with oxygen for anesthesia or euthanasia of rats. Lab Anim Sci 1997, 47:376-385.
4. Guidelines for the Euthanasia of Rodent Feti and Neonates. NIH Animal Research Advisory Committee, 2004 [http://oacu.od.nih.gov/ARAC/euthmous.pdf]
5. Klaunberg BA, O'Malley J, Clark T, Davis JA. Euthanasia of Mouse Fetuses and Neonates. Contemp Top Lab Anim Sc 2004, 43:(5) 29-34.

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